



IFWO

RAW SEQUENCE LISTING

PATENT APPLICATION: US/10/822,370

DATE: 09/16/2004

TIME: 15:26:09

Input Set : A:\BSA04-11.txt

Output Set: N:\CRF4\09162004\J822370.raw

3 <110> APPLICANT: Brookhaven Science Associates
 4 Shanklin, John
 5 Whittle, Edward J.
 7 <120> TITLE OF INVENTION: Mutant Fatty Acid Desaturase and Methods for Directed
 Mutagenesis
 9 <130> FILE REFERENCE: CIP of 10/017,145 filed December 14, 2001 which was a CIP of
 W--> 10 09/328,550 filed on June 9, 1999, which was a CIP of 09/233,856
 W--> 11 filed on January 19, 1999
 13 <140> CURRENT APPLICATION NUMBER: 10/822,370
 14 <141> CURRENT FILING DATE: 2004-04-12
 16 <150> PRIOR APPLICATION NUMBER: 09/328,550
 17 <151> PRIOR FILING DATE: 1999-06-09
 19 <150> PRIOR APPLICATION NUMBER: 10/017,145
 20 <151> PRIOR FILING DATE: 2001-12-14
 22 <150> PRIOR APPLICATION NUMBER: 09/233,856
 23 <151> PRIOR FILING DATE: 1999-01-19
 25 <160> NUMBER OF SEQ ID NOS: 19
 27 <170> SOFTWARE: PatentIn version 3.2
 29 <210> SEQ ID NO: 1
 30 <211> LENGTH: 363
 31 <212> TYPE: PRT
 32 <213> ORGANISM: Ricinus communis
 35 <220> FEATURE:
 36 <221> NAME/KEY: misc_feature
 37 <223> OTHER INFORMATION: ricinus communis delta 9 18:0 Acyl ACP Desaturase
 39 <400> SEQUENCE: 1
 41 Ala Ser Thr Leu Lys Ser Gly Ser Lys Glu Val Glu Asn Leu Lys Lys
 42 1 5 10 15
 45 Pro Phe Met Pro Pro Arg Glu Val His Val Gln Val Thr His Ser Met
 46 20 25 30
 49 Pro Pro Gln Lys Ile Glu Ile Phe Lys Ser Leu Asp Asn Trp Ala Glu
 50 35 40 45
 53 Glu Asn Ile Leu Val His Leu Lys Pro Val Glu Lys Cys Trp Gln Pro
 54 50 55 60
 57 Gln Asp Phe Leu Pro Asp Pro Ala Ser Asp Gly Phe Asp Glu Gln Val
 58 65 70 75 80
 61 Arg Glu Leu Arg Glu Arg Ala Lys Glu Ile Pro Asp Asp Tyr Phe Val
 62 85 90 95
 65 Val Leu Val Gly Asp Met Ile Thr Glu Glu Ala Leu Pro Thr Tyr Gln
 66 100 105 110
 69 Thr Met Leu Asn Thr Leu Asp Gly Val Arg Asp Glu Thr Gly Ala Ser
 70 115 120 125
 73 Pro Thr Ser Trp Ala Ile Trp Thr Arg Ala Trp Thr Ala Glu Glu Asn
 74 130 135 140



RAW SEQUENCE LISTING

DATE: 09/16/2004

PATENT APPLICATION: US/10/822,370

TIME: 15:26:09

Input Set : A:\BSA04-11.txt

Output Set: N:\CRF4\09162004\J822370.raw

```

77 Arg His Gly Asp Leu Leu Asn Lys Tyr Leu Tyr Leu Ser Gly Arg Val
78 145                150                155                160
81 Asp Met Arg Gln Ile Glu Lys Thr Ile Gln Tyr Leu Ile Gly Ser Gly
82                165                170                175
85 Met Asp Pro Arg Thr Glu Asn Ser Pro Tyr Leu Gly Phe Ile Tyr Thr
86                180                185                190
89 Ser Phe Gln Glu Arg Ala Thr Phe Ile Ser His Gly Asn Thr Ala Arg
90                195                200                205
93 Gln Ala Lys Glu His Gly Asp Ile Lys Leu Ala Gln Ile Cys Gly Thr
94                210                215                220
97 Ile Ala Ala Asp Glu Lys Arg His Glu Thr Ala Tyr Thr Lys Ile Val
98 225                230                235                240
101 Glu Lys Leu Phe Glu Ile Asp Pro Asp Gly Thr Val Leu Ala Phe Ala
102                245                250                255
105 Asp Met Met Arg Lys Lys Ile Ser Met Pro Ala His Leu Met Tyr Asp
106                260                265                270
109 Gly Arg Asp Asp Asn Leu Phe Asp His Phe Ser Ala Val Ala Gln Arg
110                275                280                285
113 Leu Gly Val Tyr Thr Ala Lys Asp Tyr Ala Asp Ile Leu Glu Phe Leu
114                290                295                300
117 Val Gly Arg Trp Lys Val Asp Lys Leu Thr Gly Leu Ser Ala Glu Gly
118 305                310                315                320
121 Gln Lys Ala Gln Asp Tyr Val Cys Arg Leu Pro Pro Arg Ile Arg Arg
122                325                330                335
125 Leu Glu Glu Arg Ala Gln Gly Arg Ala Lys Glu Ala Pro Thr Met Pro
126                340                345                350
129 Phe Ser Trp Ile Phe Asp Arg Gln Val Lys Leu
130                355                360
133 <210> SEQ ID NO: 2
134 <211> LENGTH: 1092
135 <212> TYPE: DNA
136 <213> ORGANISM: Ricinus communis
139 <220> FEATURE:
140 <221> NAME/KEY: misc_feature
141 <223> OTHER INFORMATION: residues 138 to 1239 of open reading frame
143 <400> SEQUENCE: 2
144 gcctctaccc tcaagtctgg ttctaaggaa gttgagaatc tcaagaagcc tttcatgcct      60
146 cctcgggagg tacatgttca ggttacccat tctatgccac cccaaaagat tgagatcttt      120
148 aaatccctag acaattgggc tgaggagaac attctgggtc atctgaagcc agttgagaaa      180
150 tgttggcaac cgcaggattt ttgcccagat cccgcctctg atggatttga tgagcaagtc      240
152 agggaactca gggagagagc aaaggagatt cctgatgatt attttgttgt tttggttgga      300
154 gacatgataa cggaagaagc cttccccact tatcaaacaa tgctgaatac cttggatgga      360
156 gttcgggatg aaacaggtgc aagtcctact tcttgggcaa tttggacaag ggcattggact      420
158 gcggaagaga atagacatgg tgacctctc aataagtatc tctacctatc tggacgagtg      480
160 gacatgaggc aaattgagaa gacaattcaa tatttgattg gttcaggaat ggatccacgg      540
162 acagaaaaca gtccatacct tgggttcate tatacatcat tccaggaaag ggcaaccttc      600
164 atttctcatg ggaacactgc ccgacaagcc aaagagcatg gagacataaa gttgggtcaa      660
166 atatgtggta caattgctgc agatgagaag cgccatgaga cagcctacac aaagatagtg      720
168 gaaaaactct ttgagattga tcctgatgga actggtttgg cttttgctga tatgatgaga      780

```

RAW SEQUENCE LISTING

DATE: 09/16/2004

PATENT APPLICATION: US/10/822,370

TIME: 15:26:09

Input Set : A:\BSA04-11.txt

Output Set: N:\CRF4\09162004\J822370.raw

of

```

170 aagaaaattt ctatgcctgc acacttgatg tatgatggcc gagatgataa tctttttgac      840
172 cactttttcag ctgttgcgca gcgtcttgga gtctacacag caaaggatta tgcagatata      900
174 ttggagttct tgggtggcag atggaagggtg gataaactaa cgggcctttc agctgaggga      960
176 caaaaggctc aggactatgt ttgtcggtta cctccaagaa ttagaaggct ggaagagaga     1020
178 gctcaaggaa gggcaaagga agcaccaccc atgcctttca gctggatttt cgataggcaa     1080
180 gtgaagctgt ag                                           1092
183 <210> SEQ ID NO: 3
184 <211> LENGTH: 34
185 <212> TYPE: DNA
186 <213> ORGANISM: Artificial
188 <220> FEATURE:
189 <223> OTHER INFORMATION: amplification primer
192 <220> FEATURE:
193 <221> NAME/KEY: misc_feature
194 <223> OTHER INFORMATION: PCR primer; sequence flanking unique XbaI site at the 5' end
of
195         the open reading frame
197 <400> SEQUENCE: 3
198 gtgagcggat aacaatttca cacagtctag aaat                                           34
201 <210> SEQ ID NO: 4
202 <211> LENGTH: 72
203 <212> TYPE: DNA
204 <213> ORGANISM: Artificial
206 <220> FEATURE:
207 <223> OTHER INFORMATION: amplification primer
210 <220> FEATURE:
211 <221> NAME/KEY: misc_feature
212 <222> LOCATION: (56)..(57)
213 <223> OTHER INFORMATION: PCR primer is a degenerate oligonucleotide in which "n"
indicates
214         the presence of either C, A, T or G at that nucleotide position
216 <400> SEQUENCE: 4
W--> 217 ccaaattgcc caagacgtcg gacttgcacc tgtttcatcc cgaactccat ccaamnatt      60
219 cagcattggt tg                                           72
222 <210> SEQ ID NO: 5
223 <211> LENGTH: 31
224 <212> TYPE: DNA
225 <213> ORGANISM: Artificial
227 <220> FEATURE:
228 <223> OTHER INFORMATION: amplification primer
231 <220> FEATURE:
232 <221> NAME/KEY: misc_feature
233 <223> OTHER INFORMATION: PCR primer
235 <400> SEQUENCE: 5
236 gaaacagggt caagtccgac gtcttgggca a                                           31
239 <210> SEQ ID NO: 6
240 <211> LENGTH: 26
241 <212> TYPE: DNA
242 <213> ORGANISM: Artificial
244 <220> FEATURE:
245 <223> OTHER INFORMATION: amplification primer

```

RAW SEQUENCE LISTING

DATE: 09/16/2004

PATENT APPLICATION: US/10/822,370

TIME: 15:26:09

Input Set : A:\BSA04-11.txt

Output Set: N:\CRF4\09162004\J822370.raw

248 <220> FEATURE:
 249 <221> NAME/KEY: misc_feature
 250 <223> OTHER INFORMATION: PCR primer
 252 <400> SEQUENCE: 6
 253 gttttctgtc cgcggtacca ttcctg 26
 256 <210> SEQ ID NO: 7
 257 <211> LENGTH: 34
 258 <212> TYPE: DNA
 259 <213> ORGANISM: Artificial
 261 <220> FEATURE:
 262 <223> OTHER INFORMATION: amplification primer
 265 <220> FEATURE:
 266 <221> NAME/KEY: misc_feature
 267 <223> OTHER INFORMATION: PCR primer
 269 <400> SEQUENCE: 7
 270 gtgagcggat aacaatttca cacagtctag aaat 34
 273 <210> SEQ ID NO: 8
 274 <211> LENGTH: 30
 275 <212> TYPE: DNA
 276 <213> ORGANISM: Artificial
 278 <220> FEATURE:
 279 <223> OTHER INFORMATION: amplification primer
 282 <220> FEATURE:
 283 <221> NAME/KEY: misc_feature
 284 <223> OTHER INFORMATION: PCR primer
 286 <400> SEQUENCE: 8
 287 cactgagccc ttctgtcttc aagaattctc 30
 290 <210> SEQ ID NO: 9
 291 <211> LENGTH: 28
 292 <212> TYPE: DNA
 293 <213> ORGANISM: Artificial
 295 <220> FEATURE:
 296 <223> OTHER INFORMATION: amplification primer
 299 <220> FEATURE:
 300 <221> NAME/KEY: misc_feature
 301 <223> OTHER INFORMATION: PCR primer
 303 <400> SEQUENCE: 9
 304 ttgataagtg ggaagggtt cttccgtt 28
 307 <210> SEQ ID NO: 10
 308 <211> LENGTH: 66
 309 <212> TYPE: DNA
 310 <213> ORGANISM: Artificial
 312 <220> FEATURE:
 313 <223> OTHER INFORMATION: amplification primer
 316 <220> FEATURE:
 317 <221> NAME/KEY: misc_feature
 318 <222> LOCATION: (32)-(34) *OK*
 319 <223> OTHER INFORMATION: PCR primer is degenerate oligonucleotide in which "n"
 indicates
 320 the presence of either C, A T, or G at that nucleotide position

RAW SEQUENCE LISTING

DATE: 09/16/2004

PATENT APPLICATION: US/10/822,370

TIME: 15:26:09

Input Set : A:\BSA04-11.txt

Output Set: N:\CRF4\09162004\J822370.raw

```

321      and in which "k" indicates either T or G
323 <220> FEATURE:
324 <221> NAME/KEY: misc_feature
325 <222> LOCATION: (41)..(43)
326 <223> OTHER INFORMATION: PCR primer is a degenerate oligonucleotide in which "n"
indicates
327      the presence of either C, A, T or G and in which "k" indicates
328      the presence of either T or G.
330 <220> FEATURE:
331 <221> NAME/KEY: misc_feature
332 <222> LOCATION: (44)..(46)
333 <223> OTHER INFORMATION: PCR primer is a degenerate oligonucleotide in which "n"
indicates
334      the presence of either C, A, T, or G at that nucleotide position
335      and in which "k" indicates the presence of either T or G.
337 <400> SEQUENCE: 10
W--> 338 aacggaagaa gcccttccca cttatcaaac annkctgaat npknnkgatg gagttcgga      60
340 tgaaac      66
343 <210> SEQ ID NO: 11
344 <211> LENGTH: 26
345 <212> TYPE: DNA
346 <213> ORGANISM: Artificial
348 <220> FEATURE:
349 <223> OTHER INFORMATION: amplification primer
352 <220> FEATURE:
353 <221> NAME/KEY: misc_feature
354 <223> OTHER INFORMATION: PCR primer
356 <400> SEQUENCE: 11
357 tccattcctg aaccaatcaa atattg      26
360 <210> SEQ ID NO: 12
361 <211> LENGTH: 70
362 <212> TYPE: DNA
363 <213> ORGANISM: Artificial
365 <220> FEATURE:
366 <223> OTHER INFORMATION: amplification primer
369 <220> FEATURE:
370 <221> NAME/KEY: misc_feature
371 <222> LOCATION: (22)..(24)
372 <223> OTHER INFORMATION: PCR primer in a degenerate oligonucleotide in which "n"
indicates
373      the presence of either C, A, T or G at that nucleotide position
374      and in which "k" indicates the presence of either T or G at that
375      nucleotide position.
377 <220> FEATURE:
378 <221> NAME/KEY: misc_feature
379 <222> LOCATION: (28)..(30)
380 <223> OTHER INFORMATION: PCR primer in a degenerate oligonucleotide in which "n"
indicates
381      the presence of either C, A, T or G at that nucleotide position
382      and in which "k" indicates the presence of either T or G at that
383      nucleotide position.
385 <220> FEATURE:
386 <221> NAME/KEY: misc_feature

```

RAW SEQUENCE LISTING ERROR SUMMARY
PATENT APPLICATION: US/10/822,370

DATE: 09/16/2004
TIME: 15:26:10

Input Set : A:\BSA04-11.txt
Output Set: N:\CRF4\09162004\J822370.raw

Please Note:

Use of n and/or Xaa have been detected in the Sequence Listing. Please review the Sequence Listing to ensure that a corresponding explanation is presented in the <220> to <223> fields of each sequence which presents at least one n or Xaa.

Seq#:4; N Pos. ~~56,57~~

Seq#:10; N Pos. ~~32,33,41,42,44,45~~

Seq#:12; N Pos. ~~22,23,28,29,49,50~~

Invalid <213> Response:

Use of "Artificial" only as "<213> Organism" response is incomplete, per 1.823(b) of New Sequence Rules. Valid response is Artificial Sequence.

Seq#:3,4,5,6,7,8,9,10,11,12,13,14,15,16,17,18,19

VERIFICATION SUMMARY

PATENT APPLICATION: US/10/822,370

DATE: 09/16/2004

TIME: 15:26:10

Input Set : A:\BSA04-11.txt

Output Set: N:\CRF4\09162004\J822370.raw

L:10 M:259 W: Allowed number of lines exceeded, <130> FILE REFERENCE:

L:11 M:259 W: Allowed number of lines exceeded, <130> FILE REFERENCE:

L:217 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:4 after pos.:0

L:338 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:10 after pos.:0

L:394 M:341 W: (46) "n" or "Xaa" used, for SEQ ID#:12 after pos.:0